

What is claimed is:

1. A substantially pure culture or suspension of a bacterium *Burkholderia casidae* or variant thereof, which
5 *Burkholderia casidae* or variant exhibits biocontrol activity against a microorganism, and has
 - a) a 16S rRNA gene comprising a sequence that is at least 97% similar to the sequence of SEQ ID NO:1 as determined by Clustal Analysis; and
 - 10 b) a cellular fatty acid composition comprising about 16% to about 20% C16:0 fatty acid, about 18% to about 22% C16:1 fatty acid, and about 35% to about 45% C18:1 (11,12) fatty acid;wherein the microorganism is a bacterium, yeast, filamentous
15 fungi, protozoan or algae.
2. The substantially pure culture or suspension of claim 1, which bacterium or variant thereof has a 16S rRNA gene comprising a sequence identical to that of SEQ ID NO:1;
20 and a cellular fatty acid composition comprising about 18% C16:0 fatty acid, about 21% C16:1 fatty acid and about 39% C18:1 (11,12).
3. A substantially pure culture or suspension of
25 *Burkholderia casidae* strain 2.2N having the accession number ATCC 55961, or a variant thereof.
4. The substantially pure culture or suspension of claim 1, 2 or 3, wherein the culture or suspension comprises
30 at least 80% cysts.
5. The substantially pure culture or suspension of claim 1, 2 or 3, wherein the culture or suspension comprises at least 80% cells.

35

6. The substantially pure culture or suspension of claim 1, 2 or 3, wherein the *Burkholderia casidae* or variant thereof has been inactivated.

5 7. The substantially pure culture or suspension of claim 6, wherein the *Burkholderia casidae* or variant thereof has been inactivated by treating with heat or alcohol.

8. A cell-free filtrate or cell fraction prepared from
10 the substantially pure culture or suspension of claim 1, 2 or 3.

9. A cell-free filtrate or cell fraction prepared from the substantially pure culture or suspension of claim 6.

15

10. An antimicrobial preparation comprising an alcohol-extract of a cell, cyst, culture, suspension, cell-free filtrate or cell fraction of a bacterium *Burkholderia casidae* or variant thereof, which *Burkholderia casidae* or variant
20 exhibits biocontrol activity against a microorganism, and has

a) a 16S rRNA gene comprising a sequence that is at least 97% similar to the sequence of SEQ ID NO:1 as determined by Clustal Analysis; and

b) a cellular fatty acid composition comprising about
25 16% to about 20% C16:0 fatty acid, about 18% to about 22% C16:1 fatty acid, and about 35% to about 45% C18:1 (11,12) fatty acid;

wherein the microorganism is a bacterium, yeast, filamentous fungi, protozoan or algae.

30

11. The antimicrobial preparation of claim 10, wherein the bacterium or variant thereof has a 16S rRNA gene comprising a sequence identical to that of SEQ ID NO:1; and a cellular fatty acid composition comprising about 18% C16:0
35 fatty acid, about 21% C16:1 fatty acid, and about 39% C18:1 (11,12).

12. The antimicrobial preparation of claim 11, wherein the bacterium is *Burkholderia casidae* strain 2.2N having the accession number ATCC 55961, or a variant thereof.

5 13. The antimicrobial preparation of claim 10, 11 or 12, wherein the cell, cyst, culture, suspension, cell-free filtrate or cell fraction of *Burkholderia casidae* or variant has been inactivated of any live bacterium.

10 14. The antimicrobial preparation of claim 10, 11 or 12, wherein the alcohol-extract is prepared according to a method comprising:

- 15 a) boiling an alcoholic mixture comprising the cell, culture, suspension, cell-free filtrate or cell fraction and an alcohol;
- b) clarifying the boiled mixture;
- c) mixing the boiled mixture with magnesium silicate;
- d) collecting the magnesium silicate;
- e) washing the magnesium silicate with water; and
- 20 f) eluting antifungal compounds from the magnesium silicate with an alcoholic solution, thereby producing the alcohol-extract.

 15. A biocontrol composition comprising the
25 substantially pure culture or suspension of claim 1, 2 or 3.

 16. A biocontrol composition comprising the substantially pure culture or suspension of claim 4.

30 17. A biocontrol composition comprising the substantially pure culture or suspension of claim 5.

 18. A biocontrol composition comprising the substantially pure culture or suspension of claim 6.
35

 19. A biocontrol composition comprising the substantially pure culture or suspension of claim 7.

20. A biocontrol composition comprising sprayed-dried or freeze-dried cells of the substantially pure culture or suspension of claim 1, 2 or 3.

5 21. A biocontrol composition comprising sprayed-dried or freeze-dried cells of the substantially pure culture or suspension of claim 4.

22. A biocontrol composition comprising sprayed-dried
10 or freeze-dried cells of the substantially pure culture or suspension of claim 5.

23. A biocontrol composition comprising sprayed-dried or freeze-dried cells of the substantially pure culture or
15 suspension of claim 6.

24. A biocontrol composition comprising sprayed-dried or freeze-dried cells of the substantially pure culture or suspension of claim 7.
20

25. A biocontrol composition comprising the cell-free filtrate or cell fraction of claim 8.

26. A biocontrol composition comprising the cell-free
25 filtrate or cell fraction of claim 9.

27. A biocontrol composition comprising the antimicrobial preparation of claim 10.

30 28. A biocontrol composition comprising the antimicrobial preparation of claim 11.

29. A biocontrol composition comprising the antimicrobial preparation of claim 12.
35

30. A biocontrol composition comprising the antimicrobial preparation of claim 13.

31. A biocontrol composition comprising the antimicrobial preparation of claim 14.

32. A method for treating or preventing a disease of a
5 plant, comprising applying an effective amount of the
biocontrol composition of claim 15 to the plant.

33. A method for treating or preventing a disease of a
plant, comprising applying an effective amount of the
10 biocontrol composition of claim 18 to the plant.

34. A method for treating or preventing a disease of a
plant, comprising applying an effective amount of the
biocontrol composition of claim 20 to the plant.

15

35. A method for treating or preventing a disease of a
plant, comprising applying an effective amount of the
biocontrol composition of claim 23 to the plant.

20 36. A method for treating or preventing a disease of a
plant, comprising applying an effective amount of the
biocontrol composition of claim 25 to the plant.

37. A method for treating or preventing a disease of a
25 plant, comprising applying an effective amount of the
biocontrol composition of claim 26 to the plant.

38. A method for treating or preventing a disease of a
plant, comprising applying an effective amount of the
30 biocontrol composition of claim 27 to the plant.

39. A method for treating or preventing a disease of a
plant, comprising applying an effective amount of the
biocontrol composition of claim 28 to the plant.

35

40. A method for treating or preventing a disease of a plant, comprising applying an effective amount of the biocontrol composition of claim 29 to the plant.

5 41. A method for treating or preventing a disease of a plant, comprising applying an effective amount of the biocontrol composition of claim 30 to the plant.

42. A method for treating or preventing a disease of a
10 plant, comprising applying an effective amount of the biocontrol composition of claim 31 to the plant.

43. The method according to claim 32, wherein the microorganism is a species of a genus selected from the group
15 consisting of *Alternaria*, *Aspergillus*, *Botrytis*, *Cercospora*, *Cercosporidium*, *Geotrichum*, *Mycosphaerella*, *Mucor*, *Penicillium*, *Phoma*, *Phytophthora*, *Plasmopora*, *Pseudopeziza*, *Puccinia*, *Pythium*, *Rhizoctonia*, *Rhizopus*, *Saccharomyces*, *Septoria*, *Sporothrix*, *Stemphylium*, *Trichophyton*,
20 *Verticillium*, *Erwinia*, *Pseudomonas* and *Xanthomonas*.

44. The method according to claim 33, wherein the microorganism is a species of a genus selected from the group consisting of *Alternaria*, *Aspergillus*, *Botrytis*, *Cercospora*,
25 *Cercosporidium*, *Geotrichum*, *Mycosphaerella*, *Mucor*, *Penicillium*, *Phoma*, *Phytophthora*, *Plasmopora*, *Pseudopeziza*, *Puccinia*, *Pythium*, *Rhizoctonia*, *Rhizopus*, *Saccharomyces*, *Septoria*, *Sporothrix*, *Stemphylium*, *Trichophyton*, *Verticillium*, *Erwinia*, *Pseudomonas* and *Xanthomonas*.

30

45. The method according to claim 34, wherein the microorganism is a species of a genus selected from the group consisting of *Alternaria*, *Aspergillus*, *Botrytis*, *Cercospora*, *Cercosporidium*, *Geotrichum*, *Mycosphaerella*, *Mucor*,
35 *Penicillium*, *Phoma*, *Phytophthora*, *Plasmopora*, *Pseudopeziza*, *Puccinia*, *Pythium*, *Rhizoctonia*, *Rhizopus*, *Saccharomyces*,

Septoria, Sporothrix, Stemphylium, Trichophyton, Verticillium, Erwinia, Pseudomonas and Xanthomonas.

46. The method according to claim 35, wherein the
5 microorganism is a species of a genus selected from the group
consisting of *Alternaria, Aspergillus, Botrytis, Cercospora, Cercosporidium, Geotrichum, Mycosphaerella, Mucor, Penicillium, Phoma, Phytophthora, Plasmopora, Pseudopeziza, Puccinia, Pythium, Rhizoctonia, Rhizopus, Saccharomyces,*
10 *Septoria, Sporothrix, Stemphylium, Trichophyton, Verticillium, Erwinia, Pseudomonas and Xanthomonas.*

47. The method according to claim 36, wherein the
microorganism is a species of a genus selected from the group
15 consisting of *Alternaria, Aspergillus, Botrytis, Cercospora, Cercosporidium, Geotrichum, Mycosphaerella, Mucor, Penicillium, Phoma, Phytophthora, Plasmopora, Pseudopeziza, Puccinia, Pythium, Rhizoctonia, Rhizopus, Saccharomyces, Septoria, Sporothrix, Stemphylium, Trichophyton,*
20 *Verticillium, Erwinia, Pseudomonas and Xanthomonas.*

48. The method according to claim 37, wherein the
microorganism is a species of a genus selected from the group
consisting of *Alternaria, Aspergillus, Botrytis, Cercospora,*
25 *Cercosporidium, Geotrichum, Mycosphaerella, Mucor, Penicillium, Phoma, Phytophthora, Plasmopora, Pseudopeziza, Puccinia, Pythium, Rhizoctonia, Rhizopus, Saccharomyces, Septoria, Sporothrix, Stemphylium, Trichophyton, Verticillium, Erwinia, Pseudomonas and Xanthomonas.*

30

49. The method according to claim 38, wherein the
microorganism is a species of a genus selected from the group
consisting of *Alternaria, Aspergillus, Botrytis, Cercospora, Cercosporidium, Geotrichum, Mycosphaerella, Mucor,*
35 *Penicillium, Phoma, Phytophthora, Plasmopora, Pseudopeziza, Puccinia, Pythium, Rhizoctonia, Rhizopus, Saccharomyces,*

Septoria, Sporothrix, Stemphylium, Trichophyton,
Verticillium, Erwinia, Pseudomonas and Xanthomonas.

50. The method according to claim 39, wherein the
5 microorganism is a species of a genus selected from the group
consisting of *Alternaria, Aspergillus, Botrytis, Cercospora,*
Cercosporidium, Geotrichum, Mycosphaerella, Mucor,
Penicillium, Phoma, Phytophthora, Plasmopora, Pseudopeziza,
Puccinia, Pythium, Rhizoctonia, Rhizopus, Saccharomyces,
10 *Septoria, Sporothrix, Stemphylium, Trichophyton,*
Verticillium, Erwinia, Pseudomonas and Xanthomonas.

51. The method according to claim 40, wherein the
microorganism is a species of a genus selected from the group
15 consisting of *Alternaria, Aspergillus, Botrytis, Cercospora,*
Cercosporidium, Geotrichum, Mycosphaerella, Mucor,
Penicillium, Phoma, Phytophthora, Plasmopora, Pseudopeziza,
Puccinia, Pythium, Rhizoctonia, Rhizopus, Saccharomyces,
Septoria, Sporothrix, Stemphylium, Trichophyton,
20 *Verticillium, Erwinia, Pseudomonas and Xanthomonas.*

52. The method according to claim 41, wherein the
microorganism is a species of a genus selected from the group
consisting of *Alternaria, Aspergillus, Botrytis, Cercospora,*
25 *Cercosporidium, Geotrichum, Mycosphaerella, Mucor,*
Penicillium, Phoma, Phytophthora, Plasmopora, Pseudopeziza,
Puccinia, Pythium, Rhizoctonia, Rhizopus, Saccharomyces,
Septoria, Sporothrix, Stemphylium, Trichophyton,
Verticillium, Erwinia, Pseudomonas and Xanthomonas.

30

53. The method according to claim 42, wherein the
microorganism is a species of a genus selected from the group
consisting of *Alternaria, Aspergillus, Botrytis, Cercospora,*
Cercosporidium, Geotrichum, Mycosphaerella, Mucor,
35 *Penicillium, Phoma, Phytophthora, Plasmopora, Pseudopeziza,*
Puccinia, Pythium, Rhizoctonia, Rhizopus, Saccharomyces,

Septoria, Sporothrix, Stemphylium, Trichophyton, Verticillium, Erwinia, Pseudomonas and Xanthomonas.

54. The method according to claim 32, wherein the plant
5 is selected from the group consisting of safflower, cotton,
flax, oats, canola, poinsettia, chrysanthemum, corn, soybean,
wheat, rice, alfalfa, sorghum, peanut, tobacco, tomato,
pepper, cucumber, lettuce, green bean, lima beans, peas,
cantaloupe, musk melon, citrus fruit, grape, banana,
10 geranium, azalea, rose, tulip, petunia, orchid, carnation,
pine, yew and spruce.

55. The method according to claim 33, wherein the plant
is selected from the group consisting of safflower, cotton,
15 flax, oats, canola, poinsettia, chrysanthemum, corn, soybean,
wheat, rice, alfalfa, sorghum, peanut, tobacco, tomato,
pepper, cucumber, lettuce, green bean, lima beans, peas,
cantaloupe, musk melon, citrus fruit, grape, banana,
geranium, azalea, rose, tulip, petunia, orchid, carnation,
20 pine, yew and spruce.

56. The method according to claim 34, wherein the plant
is selected from the group consisting of safflower, cotton,
flax, oats, canola, poinsettia, chrysanthemum, corn, soybean,
25 wheat, rice, alfalfa, sorghum, peanut, tobacco, tomato,
pepper, cucumber, lettuce, green bean, lima beans, peas,
cantaloupe, musk melon, citrus fruit, grape, banana,
geranium, azalea, rose, tulip, petunia, orchid, carnation,
pine, yew and spruce.

30

57. The method according to claim 35, wherein the plant
is selected from the group consisting of safflower, cotton,
flax, oats, canola, poinsettia, chrysanthemum, corn, soybean,
wheat, rice, alfalfa, sorghum, peanut, tobacco, tomato,
35 pepper, cucumber, lettuce, green bean, lima beans, peas,
cantaloupe, musk melon, citrus fruit, grape, banana,

geranium, azalea, rose, tulip, petunia, orchid, carnation,
pine, yew and spruce.

58. The method according to claim 36, wherein the plant
5 is selected from the group consisting of safflower, cotton,
flax, oats, canola, poinsettia, chrysanthemum, corn, soybean,
wheat, rice, alfalfa, sorghum, peanut, tobacco, tomato,
pepper, cucumber, lettuce, green bean, lima beans, peas,
cantaloupe, musk melon, citrus fruit, grape, banana,
10 geranium, azalea, rose, tulip, petunia, orchid, carnation,
pine, yew and spruce.

59. The method according to claim 37, wherein the plant
is selected from the group consisting of safflower, cotton,
15 flax, oats, canola, poinsettia, chrysanthemum, corn, soybean,
wheat, rice, alfalfa, sorghum, peanut, tobacco, tomato,
pepper, cucumber, lettuce, green bean, lima beans, peas,
cantaloupe, musk melon, citrus fruit, grape, banana,
geranium, azalea, rose, tulip, petunia, orchid, carnation,
20 pine, yew and spruce.

60. The method according to claim 38, wherein the plant
is selected from the group consisting of safflower, cotton,
flax, oats, canola, poinsettia, chrysanthemum, corn, soybean,
25 wheat, rice, alfalfa, sorghum, peanut, tobacco, tomato,
pepper, cucumber, lettuce, green bean, lima beans, peas,
cantaloupe, musk melon, citrus fruit, grape, banana,
geranium, azalea, rose, tulip, petunia, orchid, carnation,
pine, yew and spruce.

30

61. The method according to claim 39, wherein the plant
is selected from the group consisting of safflower, cotton,
flax, oats, canola, poinsettia, chrysanthemum, corn, soybean,
wheat, rice, alfalfa, sorghum, peanut, tobacco, tomato,
35 pepper, cucumber, lettuce, green bean, lima beans, peas,
cantaloupe, musk melon, citrus fruit, grape, banana,

geranium, azalea, rose, tulip, petunia, orchid, carnation,
pine, yew and spruce.

62. The method according to claim 40, wherein the plant
5 is selected from the group consisting of safflower, cotton,
flax, oats, canola, poinsettia, chrysanthemum, corn, soybean,
wheat, rice, alfalfa, sorghum, peanut, tobacco, tomato,
pepper, cucumber, lettuce, green bean, lima beans, peas,
cantaloupe, musk melon, citrus fruit, grape, banana,
10 geranium, azalea, rose, tulip, petunia, orchid, carnation,
pine, yew and spruce.

63. The method according to claim 41, wherein the plant
is selected from the group consisting of safflower, cotton,
15 flax, oats, canola, poinsettia, chrysanthemum, corn, soybean,
wheat, rice, alfalfa, sorghum, peanut, tobacco, tomato,
pepper, cucumber, lettuce, green bean, lima beans, peas,
cantaloupe, musk melon, citrus fruit, grape, banana,
geranium, azalea, rose, tulip, petunia, orchid, carnation,
20 pine, yew and spruce.

64. The method according to claim 42, wherein the plant
is selected from the group consisting of safflower, cotton,
flax, oats, canola, poinsettia, chrysanthemum, corn, soybean,
25 wheat, rice, alfalfa, sorghum, peanut, tobacco, tomato,
pepper, cucumber, lettuce, green bean, lima beans, peas,
cantaloupe, musk melon, citrus fruit, grape, banana,
geranium, azalea, rose, tulip, petunia, orchid, carnation,
pine, yew and spruce.

30

65. A method for producing an antimicrobial preparation
from *Burkholderia casidae* or variant thereof, comprising:
a) boiling an alcoholic mixture comprising a cell,
culture, suspension, cell-free filtrate or cell
35 fraction of *Burkholderia casidae* or variant
thereof, and an alcohol;
b) clarifying the boiled mixture;

- c) mixing the boiled mixture with magnesium silicate;
- d) collecting the magnesium silicate;
- e) washing the magnesium silicate with water; and
- f) eluting antifungal compounds from the magnesium
5 silicate with an alcoholic solution, thereby
producing the antimicrobial preparation.

66. A method for isolating a *Burkholderia casidae* or variant thereof, which comprises

- 10 (a) determining the 16S rRNA sequence and the cellular fatty acid composition of a predator bacterium which exhibits biocontrol activity against a microorganism; and
- 15 (b) identifying a bacterium having (i) a 16S rRNA gene comprising a sequence that is at least 97% similar to the sequence of SEQ ID NO:1 as determined by Clustal Analysis, and (ii) a cellular fatty acid composition comprising about 16% to about 20% C16:0 fatty acid, about 18% to about 22% C16:1 fatty
20 acid, and about 35% to about 45% C 18:1 (11,12) fatty acid;

thereby isolating the *Burkholderia casidae* or variant thereof.

- 25 67. The method of claim 66, wherein the bacterium has (i) a 16S rRNA gene comprising a sequence that is identical to the sequence of SEQ ID NO:1, and (ii) a cellular fatty acid composition comprising about 18% C16:0 fatty acid, about 21% C16:1 fatty acid and about 39% C18:1 (11,12).

30

35